

CHAPTER 74

Genetics Update: Cutaneous Malignant Melanoma and Melanocytic Nevi

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Although the first English-language report describing melanoma in 1820 contained a description of a melanoma-prone family, it was the seminal studies of investigators at the National Cancer Institute (NCI) and the University of Pennsylvania that identified dysplastic nevi as an important precursor to melanoma, suggested an autosomal dominant mode of inheritance for both melanoma and dysplastic nevi, and proposed that a melanoma susceptibility gene (*CMM1*) was located on chromosome 1p36. This gene assignment has not yet been confirmed by independent investigators. A second melanoma gene, designated *CMM2*, has been mapped to chromosome 9p21. This gene assignment has been confirmed independently, and the cell cycle regulator p16^{INK4a} has been proposed as a candidate gene, and germline mutations in this gene have been identified in about half of melanoma-prone families. Germline mutations in the cyclin-dependent kinase gene *CDK4* (chromosome 12q14) have recently been described in two melanoma kindreds, and this likely represents a third melanoma gene. A heritable determinant for total nevus number has been suggested, as has the presence of a major gene responsible for total nevus density in melanoma-prone families. Dysplastic nevi cluster in families, and an autosomal dominant mode of inheritance for dysplastic nevi, has been proposed, and evidence developed that dysplastic nevi may be a pleiotropic manifestation of the 1p36 familial melanoma gene.

INTRODUCTION

The first English-language report that described the entity we now know as “cutaneous malignant melanoma” was, in fact, a familial occurrence of the disease (1).

Familial Cancer and Prevention, Edited by Utsunomiya, Mulvihill, and Weber.
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Cawley made a similar observation in 1952 (2). Over the next 25 years, many multiple-case melanoma families were described only as interesting curiosities, prompting speculation that a hereditary form of melanoma might exist.

GENETICS OF MELANOMA

Fourteen melanoma-prone kindred were studied by investigators at the National Cancer Institute (NCI) and the University of Pennsylvania (NCI/Penn). Distinguishing features of the hereditary melanoma syndrome in the NCI/Penn series included a younger-than-average age at melanoma diagnosis, a striking predisposition toward multiple primary melanoma, and the presence of multiple, clinically atypical modes that were designated "dysplastic nevi" (3,4). In this cohort, nearly all family members with cutaneous malignant melanoma (CMM) also had dysplastic nevi (DN) on their skin and, during prospective follow-up, new melanomas were diagnosed only in family members with DN. These investigators proposed that DN were both markers that identified those family members who were at increased risk of CMM and precursor lesions from which the majority of newly diagnosed melanomas evolved. These findings were thought to be analogous to those previously made in families with colonic polyposis and colorectal cancer.

Segregation analysis suggested that when the disease trait was defined as either CMM or DN, an autosomal dominant model best fit the pattern in these families (5), a finding that has been confirmed (6). The NCI/Penn group found that the distribution of the CMM and DN was so tightly linked that they appeared to represent pleiotropic manifestations of the same gene (7). In spite of dissenting opinions from the Seventh Genetic Analysis Workshop (8), familial melanoma investigators have continued to base their analyses on the presumption that this trait is inherited in an autosomal dominant fashion.

Linkage analysis, the technique used to assign a putative genetic locus to a specific chromosomal location, was first performed by the NCI/Penn group without an a priori hypothesis as to where the melanoma gene might be. This genomic search identified moderately strong evidence of linkage between CMM/DN and the Rh blood group locus, known to be on the short arm of chromosome 1 (5). Additional analysis led to the conclusion that a CMM/DN gene is located on chromosome 1p36 (9,10). The estimated penetrance of this gene, designated *CMM1*, was 82% by age 72 (11). As yet, no candidate gene from this chromosomal region has been identified. In fact, numerous investigators have failed to corroborate the gene assignment proposed by the NCI/Penn team (12–15). Both etiologic and diagnostic heterogeneity have been suggested as explanations for this discrepancy (16). Thus, the 1p36 CMM/DN gene assignment remains viable in spite of the apparently contradictory findings of other investigators.

However, recent observations have shifted the focus of familial melanoma research to a second gene site, located on chromosome 9p. The Utah group performed a linkage analysis in 11 CMM pedigrees; DN were not included in their analysis. It provided strong evidence for a partially penetrant, dominant melanoma susceptibility

locus (designated *CMM2*) on 9p21 (17,18). The penetrance of this gene was estimated to be 53% by age 80, and gene carriers were found to have higher nevus counts and nevus densities than do nongene carriers.

The 9p21 gene assignment for the *CMM2* locus has now been confirmed (10,19–22).

The NCI group found that some of their families were linked to 9p, whereas others remained linked to 1p(10). They found statistically significant genetic heterogeneity in their cohort of families, supporting the existence of at least two melanoma-susceptibility genes and also found significant linkage to the 9p21 locus when DN were included in the analysis. Dutch investigators suggested that evidence of linkage between *CMM2* and 9p21 became stronger when DN were included in the model (19,21). British investigators evaluated six multiple-case melanoma families and found evidence supporting linkage to 9p21 in three (22).

A candidate gene for *CMM2* has been identified on chromosome 9p21. This gene is designated *p16^{INK4a}* (23). Its protein binds and inhibits cyclin-dependent kinases in vitro and is therefore thought to be an inhibitor of cell division. One could readily envision how mutations in such a protein might permit unchecked or aberrant cell proliferation in a manner similar to that attributed to tumor suppressor genes. The NCI group described germline *p16* mutations in 33 of 36 melanoma patients from 9 different families (24). These mutations were not observed in melanoma patients from families linked to the 1p36 melanoma locus, again supporting the hypothesis that are at least two melanoma susceptibility genes. The mutant p16 proteins they identified were functionally impaired in their ability to inhibit the growth-promoting activity of cyclin/cyclin-dependent kinase complexes in vitro (25). These observations provide a biochemical rationale for the hypothesis that carriers of certain *p16^{INK4a}* mutations are at increased risk of developing melanoma. However, Utah investigators analyzed *p16^{INK4a}* coding sequences in 13 families linked to 9p and in 38 additional melanoma-prone families (26). In only two families were potential predisposing mutations found. Overall, germline mutations in this gene have been identified in about half of the melanoma-prone families studied.

Goldstein et al. (27) compared the incidence of pancreatic cancer in 10 families with *p16^{INK4a}* mutations with that of 9 families with normal *p16^{INK4a}* function. There was a 22-fold increased risk of pancreatic cancer in the former (7 observed vs. 0.32 expected), while no pancreatic cancer was observed in the latter families. A report of a single family susceptible to both melanoma and pancreatic cancer with a mutation in this gene supports this observation (28). These data suggest that the development of pancreatic cancer in melanoma-prone families may require a mutation in the *p16^{INK4a}* gene.

Could there be more than two melanoma-susceptibility genes? A recent study evaluated 31 melanoma families not linked to 9p21 for evidence of mutations in other genes which are part of the cyclin-dependent kinase/cyclin D cell growth regulatory pathway. Two (6%) unrelated families were found to have the same germline mutation in *CDK4* (located on chromosome 12q14) (29). This likely represents a third melanoma gene (*CMM3*), it appears to function as a dominant oncogene, unlike most familial cancer susceptibility genes, which are tumor suppres-

sor genes. The mutated *CDK4* gene is resistant to the normal inhibition exerted by *p16^{INK4a}* and thus becomes an unregulated promoter of cell division.

GENETICS OF NEVI

The genetic basis of nevi is less well understood. With regard to nevi in general, a study of counted nevi among 23 monozygotic and 22 dizygotic twin pairs revealed a strong correlation in the total number of nevi was observed in the monozygotic twins ($r = 0.83$) but not among dizygotic twins ($r = -0.24$) (30). A precise genetic model could not be specified, but the data suggested a strong inherited basis for total nevus count.

The Utah group analyzed their families for total nevus number and total nevus density (a derived variable computed from mole size and number). Their analysis suggested the presence of a major gene that accounted for about 55% of the mole phenotype in the multiple-case families but no evidence of a major "mole gene" in the single-case families (31). Total nevus density fit a mendelian pattern better than total nevus number.

With reference to DN, the original analyses by the NCI/Penn team suggested an autosomal dominant mode of inheritance (5,9,10) and further indicated that CMM and DN might be pleiotropic manifestations of the same gene, *CMM1* (7). In a separate study, 25 patients with DN were matched to 28 controls who lacked DN, and all willing first-degree relatives of both cases and controls were examined for DN (32). DN were found among the relatives of 80% of cases and in 4% of controls. The relative risk of having DN was 7.2 if one or more relatives had DN. Three of the cases in the families multiplex for DN were found to have a first-degree relative with melanoma. This same cohort was also subjected to a formal genetic analysis (33). The estimated segregation ratio for a hypothetical DN gene was 0.52, consistent with an autosomal dominant mode of inheritance.

A skin examination was performed on 156 living family members of 31 probands initially classified as having sporadic, histologically verified DN (34). After the relatives were actually examined, 60% of the probands were found to have one or more relatives with DN. One relative was diagnosed to have malignant melanoma *in situ* at examination. Crijns et al. (34) estimated that relatives of DN probands were four times more likely than unselected patients to have DN.

British investigators examined a series of 266 melanoma patients and 305 controls for the presence of what they designated the "atypical mole syndrome" (AMS)(35), an alternative term for DN syndrome. They examined the relatives of study subjects found to have AMS, and 39% of the 91 relatives also had AMS, compared with 15% of melanoma patients and 2% of the normal population (36). Although a formal genetic analysis of nevus distribution in this cohort was not reported, Newton Bishop et al. (36) noted that the "mode of inheritance was consistent with a single autosomal dominant gene, with the AMS phenotype and melanoma as two possible expressions of the same gene."

SUMMARY

At least three genes are involved in familial melanoma. The definition of the molecular pathophysiology of two of these genes is close at hand. Heredity and familial factors are important determinants of nevus phenotype as well. The dysplastic nevus is a potent determinant of melanoma risk, both familial and nonfamilial.

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